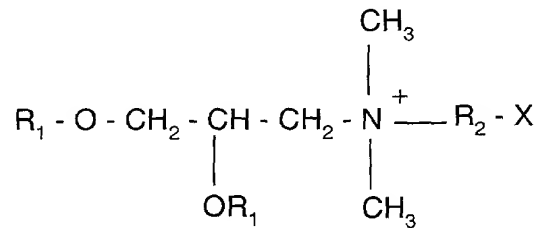


## Claims

1. DNA vaccine against a pathogen affecting farm animals, in particular bovines or porcines, comprising a plasmid containing a nucleotide sequence encoding an immunogen of a pathogen of the animal species considered, under conditions allowing the *in vivo* expression of this sequence, and a cationic lipid containing a quaternary ammonium salt, of formula



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- in which  $\text{R}_1$  is a saturated or unsaturated linear aliphatic radical having 12 to 18 carbon atoms,  $\text{R}_2$  is another aliphatic radical containing 2 or 3 carbon atoms, and X a hydroxyl or amine group, this lipid being preferably DMRIE.

2. Vaccine according to Claim 1, wherein it also comprises DOPE.

3. Vaccine according to Claim 1, wherein it comprises, in addition, a GM-CSF protein of the animal species considered.

4. Vaccine according to Claim 2, wherein it comprises, in addition, a GM-CSF protein of the animal species considered.

5. Vaccine according to Claim 1, wherein it comprises, in addition, an expression vector containing the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

6. Vaccine according to Claim 2, wherein it comprises, in addition, an expression vector containing the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

7. Vaccine according to Claim 5, wherein the expression vector is a plasmid.

8. Vaccine according to Claim 6, wherein the expression vector is a plasmid.

9. Vaccine according to Claim 1, wherein the nucleotide sequence encoding a pathogen immunogen is the sequence of a gene from which the part encoding the transmembrane domain has been deleted.

10. Vaccine according to Claims 1, wherein the plasmid containing the nucleotide sequence encoding a pathogen immunogen also contains a nucleotide sequence encoding a heterologous signal sequence, preferably a tPA.

11. Vaccine according to Claim 1, wherein the plasmid containing the nucleotide sequence encoding a pathogen immunogen also contains a stabilizing intron.

12. Vaccine according to Claim 11, wherein the intron is intron II of the rabbit beta-globin gene.

13. Vaccine according to Claim 1, wherein it comprises a nucleotide sequence of BHV-1.

14. Vaccine according to Claim 13, wherein it comprises the sequence of the gB gene optimized by a signal sequence, in particular that of the tPA signal of human origin, in place of the sequence of the signal peptide of the glycoprotein gB, and/or by the deletion of the DNA fragment encoding the transmembrane domain of gB.

15. Vaccine according to Claim 13, wherein it comprises the sequence of the gC gene optimized by a signal sequence, in particular that of the tPA signal

of human origin, in place of the sequence of the signal peptide of the glycoprotein gC, and/or by the deletion of the DNA fragment encoding the transmembrane domain of gC.

5 16. Vaccine according to Claim 13, wherein it comprises the sequence of the gD gene optimized by a signal sequence, in particular that of the tPA signal of human origin, in place of the sequence of the signal peptide of the glycoprotein gD, and/or by the deletion  
10 of the DNA fragment encoding the transmembrane domain of gD.

17. Vaccine according to Claim 13, wherein it comprises DMRIE-DOPE, an expression plasmid encoding the BHV-1 gB antigen optimized by the deletion of the  
15 fragment of the nucleotide sequence encoding the transmembrane domain and the contiguous C-terminal part, a second expression plasmid encoding the BHV-1 gC antigen optimized by the deletion of the fragment of the nucleotide sequence encoding the transmembrane  
20 domain and the contiguous C-terminal part, and a third expression plasmid encoding the BHV-1 gD antigen optimized by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain and the contiguous C-terminal part.

25 18. Vaccine according to Claim 1, wherein it comprises a nucleotide sequence of BRSV.

19. Vaccine according to Claim 18, wherein it comprises the sequence of the BRSV F gene optimized by substitution, by a signal sequence, in particular that  
30 of the tPA of human origin, of the signal sequence of the F protein of BRSV, and/or by the deletion of the DNA fragment encoding the transmembrane domain of F.

20. Vaccine according to Claim 18, wherein it comprises the sequence of the BRSV G gene optimized by

substitution, by a signal sequence, in particular that of the tPA of human origin, of the signal sequence of the G glycoprotein of BRSV, and/or by the deletion of the DNA fragment encoding the transmembrane domain  
5 of G.

21. Vaccine according to Claim 18, wherein it comprises DMRIE-DOPE, an expression plasmid encoding the F antigen of BRSV optimized by the insertion of the signal sequence of the human tPA in place of the signal  
10 sequence of F, and by the deletion of the fragment of the nucleotide sequence of F encoding the transmembrane domain and the contiguous C-terminal part, and a second expression plasmid encoding the G antigen of BRSV optimized by the insertion of the signal sequence of  
15 the human tPA in place of the signal sequence of G, and by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain of G and the contiguous C-terminal part.

22. Vaccine according to Claim 1, wherein it comprises  
20 a nucleotide sequence of BVDV.

23. Vaccine according to Claim 22, wherein it comprises the sequence of the BVDV EO gene optimized by the addition of a signal sequence, in particular that of the tPA of human origin, upstream of the nucleotide  
25 sequence encoding the EO protein, and/or by the insertion of an intron, in particular intron II of the rabbit beta-globin gene upstream of the nucleotide sequence encoding EO.

24. Vaccine according to Claim 22, wherein it  
30 comprises the sequence of the E2 gene optimized by the addition of a signal sequence, in particular that of the tPA of human origin, upstream of the nucleotide sequence encoding the E2 protein, and/or by the deletion of the DNA fragment encoding the transmembrane

domain of E2, and/or by the insertion of an intron, in particular intron II of the rabbit beta-globin gene upstream of the nucleotide sequence encoding E2.

5 25. Vaccine according to Claim 22, wherein it comprises DMRIE-DOPE, an expression plasmid encoding the EO antigen of BVDV optimized by the insertion of the signal sequence of the human tPA upstream of EO and by the insertion of intron II of the rabbit beta-globin  
10 gene upstream of EO, and a second plasmid encoding the E2 antigen of BVDV optimized by the insertion of the signal sequence of the human tPA upstream of E2, by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain of E2 and by the  
15 insertion of intron II of the rabbit beta-globin gene upstream of E2.

26. Vaccine according to Claim 1, wherein it comprises a nucleotide sequence of bPI-3.

27. Vaccine according to Claim 26, wherein it  
20 comprises the sequence of the bPI-3 HN gene optimized by substitution, by a signal sequence, in particular that of the tPA of human origin, of the signal sequence of HN, and/or by the deletion of the DNA fragment encoding the transmembrane domain of HN, and/or by the  
25 insertion of an intron, in particular of intron II of the rabbit beta-globin gene upstream of the nucleotide sequence encoding HN.

28. Vaccine according to Claim 26, wherein it  
30 comprises the sequence of the bPI-3 F gene optimized by substitution, by a signal sequence, in particular that of the tPA of human origin, of the signal sequence of F, and/or by the deletion of the DNA fragment encoding the transmembrane domain of F, and/or by the insertion of an intron, in particular of intron II of the rabbit

beta-globin gene upstream of the nucleotide sequence encoding F.

29. Vaccine according to Claim 26, wherein it comprises DMRIE-DOPE, an expression plasmid encoding the HN antigen of bPI-3 optimized by the insertion of the signal sequence of the human tPA in place of the signal sequence of HN, by the deletion of the fragment of the nucleotide sequence of HN encoding the transmembrane domain and the contiguous C-terminal part and by the insertion of intron II of the rabbit beta-globin gene upstream of HN, and a second expression plasmid encoding the F antigen of bPI-3 optimized by the insertion of the signal sequence of the human tPA in place of the signal sequence of F, by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain of F and the contiguous C-terminal part and by the insertion of intron II of the rabbit beta-globin gene upstream of F.

30. Vaccine according to Claim 1, wherein it comprises a nucleotide sequence of PRV.

31. Vaccine according to Claim 30, wherein it comprises the sequence of the gB gene optimized by substitution, by a signal sequence, in particular that of the tPA signal of human origin, of the sequence of the signal peptide of the gB glycoprotein, and/or by the deletion of the DNA fragment encoding the transmembrane domain of gB.

32. Vaccine according to Claim 30, wherein it comprises the sequence of the gC gene optimized by substitution, by a signal sequence, in particular that of the tPA signal of human origin, of the sequence of the signal peptide of the gC glycoprotein, and/or by the deletion of the DNA fragment encoding the transmembrane domain of gC.

33. Vaccine according to Claim 30, wherein it comprises the sequence of the gD gene optimized by substitution, by a signal sequence, in particular that of the tPA signal of human origin, of the sequence of the signal peptide of the gD glycoprotein, and/or by the deletion of the DNA fragment encoding the transmembrane domain of gD.

34. Vaccine according to Claim 30, wherein it comprises DMRIE-DOPE, an expression plasmid encoding the gB antigen of PRV optimized by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain and of the contiguous C-terminal part, a second expression plasmid encoding the gC antigen of PRV optimized by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain and of the contiguous C-terminal part, and a third expression plasmid encoding the gD antigen of PRV optimized by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain and of the contiguous C-terminal part.

35. Vaccine according to Claim 1, wherein it comprises a nucleotide sequence of PRRSV.

36. Vaccine according to Claim 35, wherein it comprises a nucleotide sequence of the ORF3 gene optimized by substitution, by a signal sequence, in particular that of the tPA signal of human origin, or the sequence of the signal peptide of the protein encoded by ORF3, and/or by the deletion of the DNA fragment encoding the transmembrane domain of ORF3.

37. Vaccine according to Claim 35, wherein it comprises a nucleotide sequence of the ORF5 gene optimized by substitution, by a signal sequence, in particular that of the tPA signal of human origin, or

the sequence of the signal peptide of the protein encoded by ORF5, and/or by the deletion of the DNA fragment encoding the transmembrane domain of ORF5.

38. Vaccine according to Claim 35, wherein it  
5 comprises a nucleotide sequence of the ORF6 gene optimized by substitution, by a signal sequence, in particular that of the tPA signal of human origin, or the sequence of the signal peptide of the protein encoded by ORF6, and/or by the deletion of the DNA  
10 fragment encoding the transmembrane domain of ORF6.

39. Vaccine according to Claim 35, wherein it comprises DMR1E-DOPE, an expression plasmid encoding the ORF3 antigen of PRRSV, a second expression plasmid encoding the ORF5 antigen of PRRSV optimized by  
15 substitution of the signal sequence of ORF5 by the human tPA signal peptide sequence and by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain and the contiguous C-terminal part, and a third expression plasmid encoding the ORF6  
20 antigen of PRRSV optimized by the substitution of the signal sequence of ORF6 by the human tPA signal peptide sequence and by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain and the contiguous C-terminal part.

25 40. Vaccine according to Claim 1, wherein it comprises a nucleotide sequence of SIV.

41. Vaccine according to Claim 40, wherein it comprises a nucleotide sequence of the HA gene optimized by substitution, by a signal sequence, in  
30 particular that of the tPA of human origin, of the signal sequence of HA, and/or by the deletion of the DNA fragment encoding the transmembrane domain of HA, and/or by the insertion of an intron, in particular of



intron II of the rabbit beta-globin gene upstream of the nucleotide sequence encoding HA.

42. Vaccine according to Claim 40, wherein it comprises a nucleotide sequence of the NA gene optimized by substitution, by a signal sequence, in particular that of the tPA of human origin, of the signal sequence of NA, and/or by the deletion of the DNA fragment encoding the transmembrane domain of NA, and/or by the insertion of an intron, in particular of intron II of the rabbit beta-globin gene upstream of the nucleotide sequence encoding NA.

43. Vaccine according to Claim 40, wherein it comprises DMRIE-DOPE, an expression plasmid encoding the HA antigen of SIV optimized by the insertion of the signal sequence of the human tPA in place of the signal sequence of HA, by the deletion of the fragment of the nucleotide sequence of HA encoding the transmembrane domain and the contiguous C-terminal part, and by the insertion of intron II of the rabbit beta-globin gene upstream of HA, and a second expression plasmid encoding the NA antigen of SIV optimized by the insertion of the signal sequence of the human tPA in place of the signal sequence of NA, by the deletion of the fragment of the nucleotide sequence encoding the transmembrane domain of NA and the contiguous C-terminal part, and by the insertion of intron II of the rabbit beta-globin gene upstream of NA.

44. Vaccine according to claim 9, wherein it also comprises DOPE.

45. Vaccine according to claim 9, wherein it comprises, in addition, a GM-CSF protein of the animal species considered.

46. Vaccine according to claims 9, wherein it comprises, in addition, an expression vector containing

the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

47. Vaccine according to claim 9, wherein the  
5 expression vector is a plasmid.

48. Vaccine according to claim 10, wherein it also comprises DOPE.

49. Vaccine according to claim 10, wherein it comprises, in addition, a GM-CSF protein of the animal  
10 species considered.

50. Vaccine according to claim 10, wherein it comprises, in addition, an expression vector containing the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in*  
15 *vivo* expression of this sequence.

51. Vaccine according to claim 10, wherein the expression vector is a plasmid.

52. Vaccine according to claim 11, wherein it also comprises DOPE.

53. Vaccine according to claim 11, wherein it comprises, in addition, a GM-CSF protein of the animal  
20 species considered.

54. Vaccine according to claim 11, wherein it comprises, in addition, an expression vector containing  
25 the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

55. Vaccine according to claim 11, wherein the expression vector is a plasmid.

56. Vaccine according to claim 13, wherein it also comprises DOPE.

57. Vaccine according to claim 13, wherein it comprises, in addition, a GM-CSF protein of the animal species considered.

58. Vaccine according to claim 13, wherein it comprises, in addition, an expression vector containing the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

59. Vaccine according to claim 13, wherein the expression vector is a plasmid.

60. Vaccine according to claim 18, wherein it also comprises DOPE.

10 61. Vaccine according to claim 18, wherein it comprises, in addition, a GM-CSF protein of the animal species considered.

62. Vaccine according to claim 18, wherein it comprises, in addition, an expression vector containing  
15 the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

63. Vaccine according to claim 18, wherein the expression vector is a plasmid.

20 64. Vaccine according to claim 22, wherein it also comprises DOPE.

65. Vaccine according to claim 22, wherein it comprises, in addition, a GM-CSF protein of the animal species considered.

25 66. Vaccine according to claim 22, wherein it comprises, in addition, an expression vector containing the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

30 67. Vaccine according to claim 22, wherein the expression vector is a plasmid.

68. Vaccine according to claim 26, wherein it also comprises DOPE.

69. Vaccine according to claim 26, wherein it comprises, in addition, a GM-CSF protein of the animal species considered.

70. Vaccine according to claim 26, wherein it  
5 comprises, in addition, an expression vector containing the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

71. Vaccine according to claim 26, wherein the  
10 expression vector is a plasmid.

72. Vaccine according to claim 30, wherein it also comprises DOPE.

73. Vaccine according to claim 30, wherein it  
15 comprises, in addition, a GM-CSF protein of the animal species considered.

74. Vaccine according to claim 30, wherein it  
20 comprises, in addition, an expression vector containing the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

75. Vaccine according to claim 30, wherein the expression vector is a plasmid.

76. Vaccine according to claim 35, wherein it also comprises DOPE.

25 77. Vaccine according to claim 35, wherein it comprises, in addition, a GM-CSF protein of the animal species considered.

78. Vaccine according to claim 35, wherein it  
30 comprises, in addition, an expression vector containing the gene encoding the GM-CSF protein of the animal species considered, under conditions allowing the *in vivo* expression of this sequence.

79. Vaccine according to claim 35, wherein the expression vector is a plasmid.

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